Increased Dopamine Transmission in Schizophrenia: Relationship to Illness Phases

Marc Laruelle, Anissa Abi-Dargham, Roberto Gil, Lawrence Kegeles, and Robert Innis

Background: *Abnormalities of dopamine function in schizophrenia are suggested by the common antidopaminergic properties of antipsychotic medications. However, direct evidence of a hyperdopaminergic state in schizophrenia has been difficult to demonstrate, given the difficulty to measure dopamine transmission in the living human brain. Such evidence has recently emerged. Three studies reported an increase in dopamine transmission following acute amphetamine challenge in patients with schizophrenia compared to matched healthy control subjects, thus demonstrating a dysregulation of dopamine in schizophrenia. In all studies, a large variance was observed within the schizophrenic group in the magnitude of this finding, and clinical predictors of this effect could not be identified.*

Methods: *In this paper, we combined previously published and newly acquired data to obtain sufficient power to address this question.*

Results: *The most important findings derived from this extended data set are: 1) dysregulation of dopamine function revealed by the amphetamine challenge is present at onset of illness and in patients never previously exposed to neuroleptic medications; 2) this dysregulation was observed in patients experiencing an episode of illness exacerbation, but not in patients studied during a remission phase.*

Conclusions: *A hyperdopaminergic state is present in schizophrenia during the initial episode and subsequent relapses, but not in periods of remission. This finding has important consequences for the development of new treatment strategies for the remission phase.* Biol Psychiatry 1999;46:56–72 © *1999 Society of Biological Psychiatry*

Key Words: Schizophrenia, dopamine, D_2 receptors, SPECT, psychostimulants

Introduction

The "classical" dopamine hypothesis of schizophrenia proposes that hyperactivity of dopamine transmission is responsible for positive symptoms of the disorder (Carlsson and Lindqvist 1963). This hypothesis was supported by the correlation between clinical doses of antipsychotic drugs and their potency to block dopamine D_2 receptors (Creese et al 1976; Seeman and Lee 1975), and by the psychotogenic effects of dopamine enhancing drugs (for review see Angrist and van Kammen 1984; Lieberman et al 1987a). Since positive symptoms are more sensitive than negative symptoms to direct manipulation of the dopamine system, hyperactivity of dopamine transmission is likely to be more relevant to positive than negative symptoms (Crow 1980). These pharmacologic effects suggest, but do not establish, a dysregulation of dopamine systems in schizophrenia.

Despite decades of effort to validate this hypothesis, documentation of abnormalities of dopamine function in schizophrenia has remained elusive. Postmortem studies measuring dopamine and its metabolites in the brain of schizophrenic patients have yielded inconsistent results (for review see Davis et al 1991). Increased density of striatal D_2 and D_2-like receptors, reported in most postmortem studies (Lee et al 1978; Owen et al 1978; for review see Seeman et al 1987), has been difficult to interpret, given that neuroleptic drugs upregulate these receptors (Burt et al 1977; Seeman 1987). PET and SPECT studies of striatal D_2 and D_2-like receptor density in neuroleptic-naive patients with schizophrenia have generally been negative (Breier et al 1997; Farde et al 1990; Hietala et al 1994; Laruelle et al 1996; Pilowsky et al 1994), but see Wong and co-workers (1986). A recent meta-analysis of 13 in vivo studies revealed that D_2 receptor density might be elevated in schizophrenia, but the effect size was small (.54) (Laruelle 1998). The lack of clear evidence for increased dopaminergic indices in schizophrenia might indicate that dopamine transmission is enhanced only relative to other systems, such as the glutamatergic system (Carlsson 1988; Hietala and Syvalahti 1996; Willner 1997). On the other hand, the

From the Department of Psychiatry, Columbia University College of Physicians and Surgeons and New York State Psychiatric Institute, New York, NY (ML, AA-D, RG, LK); and the Department of Psychiatry, Yale University School of Medicine, New Haven, and Veterans Administration Connecticut Health Care System, West Haven, CT (ML, AA-D, RG, RI).

Address reprint requests to Marc Laruelle, MD, Department of Psychiatry, Columbia University, New York State Psychiatric Institute, 1051 Riverside Drive, Unit 28, New York, NY, 10032.

Received November 10, 1998; revised March 4, 1999; accepted March 8, 1999.

0006-3223/99/$20.00
PII S0006-3223(99)00067-0

absence of data supporting the dopamine hypothesis of schizophrenia might be due to the difficulty in obtaining a direct measurement of dopamine transmission in the living human brain.

Several groups have recently provided evidence that competition between endogenous levels of dopamine and radioligands for binding to D_2 receptors allows measurement of changes in synaptic dopamine levels with in vivo binding techniques. These interactions have been demonstrated in rodents (Inoue et al 1991; Köhler et al 1981; Ross 1991; Ross and Jackson 1989a; Ross and Jackson 1989b; Seeman et al 1989a; Van der Werf et al 1986; Young et al 1991), nonhuman primates (Carson et al 1997; Dewey et al 1993; Innis et al 1992; Laruelle et al 1997b; Logan et al 1991; Mukherjee et al 1997), and humans (Booij et al 1997; Farde et al 1992; Laruelle et al 1995; Volkow et al 1994).

In 1996, we reported that amphetamine (0.3 mg/kg, IV) induced a larger displacement of the SPECT D_2/D_3 antagonist [^{123}I]IBZM in a group of 15 schizophrenic patients compared to 15 healthy control subjects (Laruelle et al 1996). This result was independently confirmed by Breier and co-workers (1997) using PET, [^{11}C]raclopride and a lower dose of amphetamine (0.2 mg/kg) in a group of 11 patients with schizophrenia and 11 control subjects. We replicated this result in a new cohort of 15 patients and 15 control subjects (Abi-Dargham et al 1998). Together, these reports indicate that schizophrenia is associated with a larger stimulation of D_2 receptor transmission following amphetamine challenge compared to healthy control subjects. Providing that the affinity of D_2 receptors for dopamine is unchanged in schizophrenia (an assumption that remains to be firmly established in vivo), these findings are best explained by a larger increase in intrasynaptic dopamine following amphetamine challenge in patients with schizophrenia.

Moreover, the data also indicated that this regulation had a clinical significance. In all three studies, a correlation was found between the exaggerated response of the dopaminergic system and a transient worsening of the patients' symptomatology. This correlation was mostly noted with the exacerbation of positive symptoms, documenting the role of dopamine in their expression.

In all studies, a large variance was observed within the patients groups in the amphetamine effects on dopamine transmission. The limited number of cases in each cohort precluded the identification of clinical factors that could be associated with increased amphetamine effect. Another remaining question was the role of previous neuroleptic treatment. While all patients were drug free at the time of the study, almost all of them had received considerable previous exposure to D_2 receptor antagonists, and the role of previous antipsychotic treatment in this exaggerated dopaminergic response was unclear. Another unanswered question was whether this dopaminergic abnormality was present in both male and female patients. The previously published studies were performed in a Veterans Administration medical center, and the small number of female patients in each study precluded the analysis of a potential gender effect.

Table 1. Demographic Composition of the Groups

	Controls	Patients with Schizophrenia	*p*
n	36	34	
Age	40 ± 9	40 ± 9	
Gender (M/F)	32/4	28/6	
Race (C/AA/H)	21/10/3	18/11/5	.95
Parental SES	34.3 ± 9.7	34.4 ± 15.8	.96
Subject SES	39.5 ± 12.1	26.0 ± 10.2	<.001

M, male; F, female; C, Caucasian; AA, African American; H, Hispanic; SES, Socioeconomic status, measured with Hollingshead scale (Hollingshead, 1975).

In this report, we combine the results of our two previously published cohorts (Abi-Dargham et al 1998; Laruelle et al 1996), and we add the results of 10 new experiments (six healthy control subjects and four first episode, drug-naive patients) recently acquired. Thus, this report includes 36 control subjects and 34 patients, of whom 7 are first episode, drug-naive patients. The goal of this analysis was to compare in this larger sample, drug-naive and previously treated patients, to test the effect of gender, and to try to identify clinical features associated with more prominent dysregulation of the dopamine system.

Methods and Materials

Subjects

A total of 70 subjects were included in this analysis (36 normal control subjects and 34 patients with schizophrenia) (Table 1). The sample includes 15 patients and 15 control subjects from our first cohort (Laruelle et al 1996), 15 patients and 15 control subjects from our second cohort (Abi-Dargham et al 1998), and a third cohort of 6 control subjects and 4 patients not previously reported. The first and second cohorts were acquired at Yale University, while the third cohort was acquired at Columbia University. Since all cohorts were acquired with the same experimental protocol, the same type of SPECT camera (PRISM 3000, Picker, OH), and under the direction of the same investigators (ML, AA, and RG), the results could be pooled for the present analysis. This assumption was validated by testing the existence of a cohort or site effect in the control samples.

Inclusion criteria for patients were as follows: 1) diagnosis of schizophrenia according to DSM-IV; 2) no other DSM-IV Axis I diagnosis; 3) no history of alcohol or substance abuse or dependence; 4) absence of any psychotropic medications for at least 21 days prior to the study (with the exception of lorazepam, which was allowed at a maximal dose of 3 mg per day up to 24 h prior to the study); 5) no concomitant or past severe medical

conditions; 6) no pregnancy; 7) no current suicidal or homicidal ideation; and 8) ability to provide informed consent.

The studies presented in this paper were approved by four institutional review boards (IRB). Studies conducted at Yale were approved by the Yale University IRB and West Haven Veteran Administration Medical Center IRB. Studies conducted at Columbia were approved by the New York State Psychiatric Institute IRB and the Columbia Presbyterian Medical Center IRB. Patients provided informed consent after detailed explanation of the nature and risks of the study. The ability of the patient to provide informed consent was formally evaluated by a psychiatrist not associated with the study. According to the recommendations of the National Alliance for the Mentally Ill (Arlington, VA), assent of involved family members was also obtained.

All patients were admitted to an inpatient unit for the duration of the study, including the washout period. Patients were recruited under two modalities. A first group of 17 patients were recruited shortly after admission to the hospital for clinical reasons (West Haven VA for the Yale sample, and New York State Psychiatric Institute, Schizophrenia Research Unit for the Columbia sample). In all cases, the admission was voluntary. The second group of 17 patients were recruited from outpatient clinics during a stable phase of the illness, and admitted to the hospital for the purpose of the study (elective admission).

In the patients group, 7 subjects were neuroleptic naive and experiencing a first episode of the illness. Among the chronic patients, 9 patients were taking neuroleptics and/or other psychotropic drugs at the time of recruitment. In these patients, the duration of neuroleptic washout was 29 ± 6 days. Eighteen patients were neuroleptic free at the time of recruitment for reasons unrelated to the study (such as noncompliance or intolerance). In these patients, the average neuroleptic free interval was 142 ± 130 days (using an index of 365 days for patients drug free since more than 1 year, $n = 4$). Combining all previously treated patients, the average neuroleptic-free interval was 104 ± 118 days.

Inclusion criteria for control subjects were 1) absence of past or present neurologic or psychiatric illnesses, including history of substance or alcohol abuse and dependence (as assessed by interview and negative urine toxicology); 2) no concomitant or past severe medical conditions; 3) no pregnancy; and 4) informed consent. Groups were matched for age, gender, race, and parental socioeconomic level as assessed by the Hollingshead scale (Hollingshead 1975) (Table 1). The socioeconomic status of the patients was significantly lower than control subjects (Table 1).

Scan Protocol

SPECT experiments were carried out as previously described (Laruelle et al 1995). Briefly, [^{123}I]IBZM with specific activity >5000 Ci/mmol and radiochemical purity >95% was prepared by direct electrophilic radioiodination of the desiodoprecursor BZM. A total [^{123}I]IBZM dose of 10.3 ± 2.5 mCi (with these and subsequent values expressed as mean ± SD) was given as a bolus followed by a continuous infusion for the duration of the experiment (360 min). The activity given as a bolus was 3.9-fold higher than the activity infused per hour. This protocol of administration (bolus plus constant infusion with bolus to hourly infusion ratio of 3.9 hours) has been shown to induce a state of sustained binding equilibrium. In the absence of amphetamine injection, both the specific and nonspecific activity remained at a constant level (within ± 5%) from 150 min to the end of the experiment (Laruelle et al 1995).

SPECT data were acquired on the PRISM 3000 (Picker, Cleveland, OH) with high resolution fan beam collimators (resolution at full width half-maximum, 11 mm; ^{123}I point source sensitivity, 16.5 counts/sec/μCi). Two scanning sessions were obtained for each subject during the course of the [^{123}I]IBZM infusion (before and after amphetamine injection). Each scanning session lasted 60 min. The first scanning session was obtained from 180 to 240 min, followed by an injection of dextroamphetamine sulfate IV at a dose of 0.3 mg/kg over 30 sec. During the 60 min following the amphetamine injection, subjects were outside the scanner to facilitate the evaluation of the physiologic and psychiatric response to amphetamine. The second scanning session (i.e., post-amphetamine) was obtained from 300 to 360 min.

Plasma metabolite-corrected [^{123}I]IBZM steady-state concentration (C_{SS}) was measured by extraction followed by high-pressure liquid chromatography on four venous samples collected at 20 min intervals from 180 to 260 min (Laruelle et al 1995). Determination of the plasma [^{123}I]IBZM free fraction (f_1) was performed by ultrafiltration (Centrifree, Amicon, Danvers, MA) (Gandelman et al 1994). Amphetamine plasma concentration was measured by gas chromatography (National Medical Services Inc., Willow Grove, PA) on three venous samples obtained at 10, 20, and 40 min post-amphetamine injection.

No statistically significant differences were observed between the groups in experimental parameters such as injected dose, timing of the different phases of the experiment, or time of day at which experiments were performed. Patients and control subjects were acquired in parallel, to correct for potential seasonal effects. Data were acquired between September 1994 and September 1998.

Evaluation of Clinical Response

The clinical response to the amphetamine challenge was evaluated with the Positive and Negative Symptom Scale (PANSS) (Kay et al 1987). Baseline ratings were obtained 60 min before the first scanning session. Post-amphetamine ratings were obtained 30 min after the injection of amphetamine (i.e., during the interval between the first and second scanning sessions). The PANSS positive and negative subscales include seven items, each scored from 1 (not present) to 7 (extreme). Thus, the total PANSS positive and negative subscales vary from 7 (minimum) to 49 (maximum), i.e., a range of 42 points. For each of these subscales, a change of at least 4 points relative to baseline was considered clinically significant. This threshold (change of 4 points in a 42-range scale) was used to denote a clinically noticeable change, and not necessarily a clinically severe change. Behavioral response was also evaluated by the subjects with a simplified version of the Amphetamine Interview Rating Scale (van Kammen and Murphy 1975). Four items were rated on a scale from 0 (not at all) to 10 (most ever) at various intervals

before and after the amphetamine injection: euphoria ("feel good"), alertness ("feel energetic"), restlessness ("feel like moving"), and anxiety ("feel anxious").

Data Analysis

SPECT images were analyzed as previously described (Laruelle et al 1996). The baseline [^{123}I]IBZM binding potential (mL g^{-1}), corresponding to the product of the free receptor density (B_{max}, nmol/L or pmol per g of brain tissue) and affinity ($1/K_D$, nmol/L^{-1}, or mL of plasma per pmol), was calculated as the ratio of striatal specific binding (μCi per g of brain tissue) to the steady-state free unmetabolized plasma tracer concentration (f_1C_{SS}, μCi per mL of plasma) measured during scanning session 1. For each scanning session, the specific to nonspecific equilibrium partition coefficient (V_3") was calculated as the ratio of striatal minus nonspecific to nonspecific activity. Under steady-state conditions, the decrease in specific to nonspecific partition coefficient is equivalent to the decrease in binding potential (Laruelle et al 1995). Amphetamine-induced decrease in [^{123}I]IBZM binding potential was expressed in percentage of pre-amphetamine value.

Unless otherwise specified, between-group comparisons were performed with ANOVA, followed by post-hoc Fisher's PLSD. Relationships between continuous variables were analyzed with the Pearson product moment correlation coefficient. A probability value of .05 was selected as significance level.

Results

Group Comparison

In control subjects, amphetamine-induced reduction in [^{123}I]IBZM binding potential was 7.5 ± 7.1% (n = 36). The amphetamine effect was similar across the three cohorts of healthy controls, supporting the consistency of the protocol and procedures (7.6 ± 8.0%, 7.1 ± 6.3%, and 8.3 ± 7.7% for control cohorts 1, 2, and 3, respectively; p = .94). Compared to control subjects, patients with schizophrenia displayed a marked elevation of amphetamine-induced [^{123}I]IBZM displacement (17.1 ± 13.2%, p = .0003, Table 2, Figure 1). The effect size of the difference, calculated as the difference of the means divided by the average SD, was .95. The variance was larger in the schizophrenic group compared to the control group (variance ratio: 3.49, p = .004). Because of the difference in variance, we also compared the groups with a nonparametric test, and obtained the same results (Mann–Whitney, p = .0028).

This increased effect of amphetamine in patients with schizophrenia was not related to differences in amphetamine plasma disposition, since amphetamine plasma levels were similar in both groups (controls: 27.7 ± 11.0 ng/mL; patients with schizophrenia: 28.7 ± 9.9 ng/mL; p = .73). Moreover, no relationship was found between amphetamine plasma levels and [^{123}I]IBZM displacement, either in the controls (r = .04, p = .84), or in the patient group (r = .05, p = .80).

Table 2. Results: Comparison of Control Subjects and Patients with Schizophrenia

Outcome Measure	Control Subjects	Patients with Schizophrenia	p
[^{123}I]IBZM BP (mL/g) (dopamine D_2 receptors)	217 ± 63	224 ± 92	.76
Amphetamine-induced relative decrease in [^{123}I]IBZM BP (% baseline)	7.5% ± 7.1%	17.1% ± 13.2%	<.001
Amphetamine plasma concentration (ng/mL)	28 ± 11	29 ± 10	.74

Correlation With Changes in Positive Symptoms

In patients with schizophrenia, the amphetamine challenge induced an increase in positive symptoms (the positive symptoms subscale of the PANSS increased from 17.5 ± 6.2 to 20.5 ± 7.6, repeated measures ANOVA, p = .019). A large between subject variability was observed in the amphetamine-induced changes in positive symptoms (range from −8 to +13). Using the criteria of a change of 4 points in the PANSS positive subscale as the

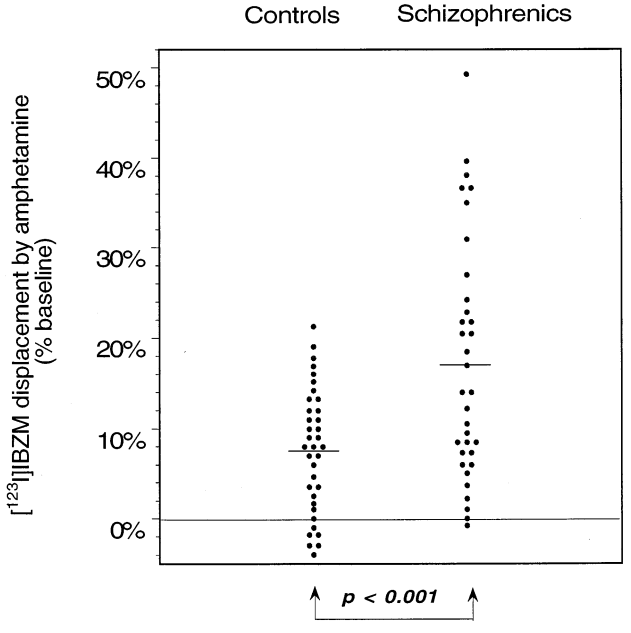

Figure 1. Effect of amphetamine (0.3 mg/kg) on [^{123}I]IBZM binding in healthy control subjects and untreated patients with schizophrenia. The y-axis shows the percentage decrease in [^{123}I]IBZM binding potential induced by amphetamine, which is a measure of the increased occupancy of D_2 receptors by dopamine following the challenge.

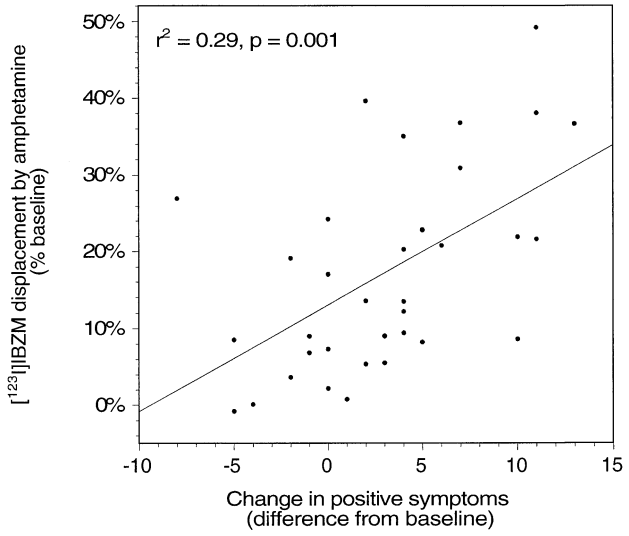

Figure 2. Relationship between striatal amphetamine-induced dopamine release and amphetamine-induced changes in positive symptoms.

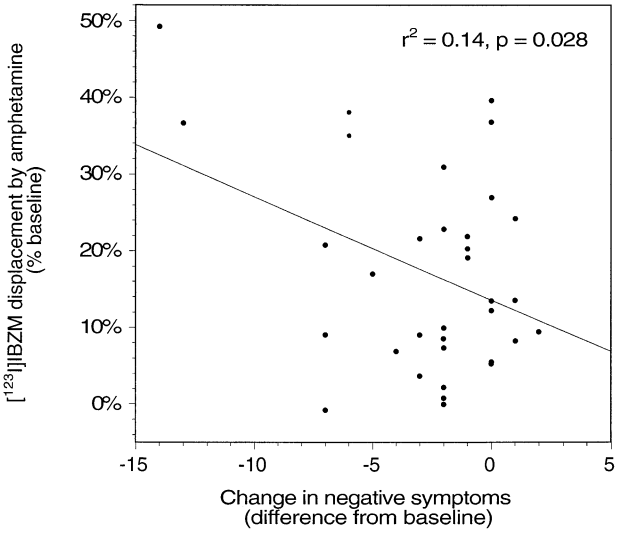

Figure 3. Relationship between striatal amphetamine-induced dopamine release and amphetamine-induced changes in negative symptoms.

threshold of clinical significance, 16 of the 34 patients (47% of the sample) were described as "worseners" with positive symptoms, 14 (41%) were described as "no change," and 4 (12%) as "improvers." The severity of positive symptoms at baseline was not associated with the magnitude of the change in positive symptoms induced by the challenge ($r = .10$, $p = .54$).

Contrasting the worseners in positive symptoms with the nonworseners (i.e., no change or improvers), patients who worsened in positive symptoms displayed an [^{123}I]IBZM displacement of 24.1 ± 12.4% ($n = 16$), while patients who did not worsen with positive symptoms showed only 10.9 ± 10.8% [^{123}I]IBZM displacement ($n = 18$), and this difference was statistically significant ($p = .0022$). Moreover, we observed a significant correlation between the increase in positive symptoms and the [^{123}I]IBZM displacement ($r = .54$, $p = .0009$, Figure 2). The severity of positive symptoms at baseline was not predictive of the magnitude of [^{123}I]IBZM displacement ($r < .01$, $p = .98$).

The emergence or worsening of positive symptoms was transient, and patients returned to their baseline symptomatology within a few hours of the challenge. No emergency medication was needed to control these symptoms.

Correlation With Changes in Negative Symptoms

The amphetamine challenge also resulted in a transient improvement in negative symptoms, with the negative subscale of the PANSS decreasing from 16.8 ± 6.6 to 14.1 ± 5.8 (repeated measures ANOVA, $p = .0001$). The severity of negative symptoms at baseline was predictive of the magnitude of improvement in negative symptoms induced by amphetamine: the more severe the negative symptoms, the more likely were they to improve following amphetamine ($r = .48$, $p = .004$). Using the same criteria of clinical significance used for positive symptoms (change of at least 4 points on the negative subscale), 25 patients qualified as nonimprovers (74% of the sample) and 9 as improvers on negative symptoms (26% of the sample). We observed a trend for patients whose negative symptoms improved to display larger [^{123}I]IBZM displacement (23.6 ± 17.0%) than patients whose negative symptoms did not improve (14.8 ± 11.1%, $p = .089$), and a significant correlation between improvement in negative symptoms and [^{123}I]IBZM displacement ($r = 0.37$, $p = .028$, Figure 3). However, as shown in Figure 3, this correlation was mostly driven by two patients who manifested a marked improvement in negative symptoms and a large [^{123}I]IBZM displacement. After removing these two patients, no correlation was observed between improvement in negative symptoms and [^{123}I]IBZM displacement ($r = .01$, $p = .98$). The severity of baseline negative symptoms was not predictive of the [^{123}I]IBZM displacement ($r = .03$, $p = .84$).

Effect of Procedure-Induced Stress

Since stress is known to stimulate dopamine release, we examined the contribution of the procedure related stress, as experienced by the subject, to the variance of the dopaminergic response. We used the self-reports of anxiety as a subjective measure of stress. First, patients with schizophrenia were significantly more anxious (4.2 ± 1.9)

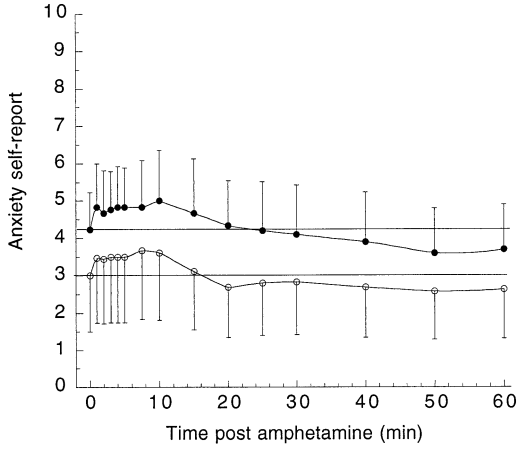

Figure 4. Amphetamine-induced changes in self-report of anxiety in patients with schizophrenia (closed circle, mean ± SD) and control subjects (open circles). Solid lines represent baseline (pre-amphetamine) anxiety scores for patients and control subjects. Amphetamine induced an initial increase in anxiety, which peaked at about 10 min in both groups. Following this initial increase, anxiety decreased below baseline levels in both groups.

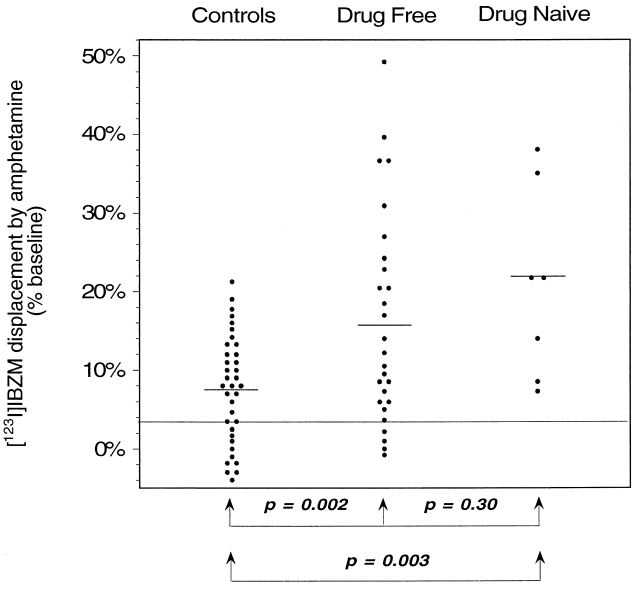

Figure 5. Amphetamine-induced reduction in [^{123}I]IBZM binding potential in healthy control subjects, chronic schizophrenic patients previously exposed to antipsychotic drugs, and in first-episode patients, never previously exposed to antipsychotic drugs.

prior to the amphetamine injection than control subjects (3.0 ± 1.9, $p = .012$). Second, the increase over baseline in anxiety produced by the amphetamine challenge was moderate, and not different between groups (patients: $+2.3 \pm 2.0$; controls: $+1.9 \pm 2.1$, $p = .41$) (Figure 4). Given their higher baseline value, the peak anxiety score was higher in patients (6.5 ± 2.4) than controls (4.9 ± 2.5, $p = .001$). The peak in anxiety score was measured at about 10 min. Following this peak, anxiety scores decreased to baseline levels in both groups. In fact, we observed a trend for anxiety self-reports to be lower at 60 min post-amphetamine than before amphetamine, with no group by time interaction (repeated measures ANOVA, time factor, $p = .07$; group factor, $p = .007$; interaction, $p = .70$).

Thus, patients experienced more anxiety than controls, both before and during the challenge. However, none of the three anxiety measures (baseline, increase over baseline, or peak) was correlated with the [^{123}I]IBZM displacement, and this absence of correlation was noted both in the control subjects ($r = .17$, .01, and .14, respectively) and in the patients ($r = .12$, .02, and .22, respectively). In patients, the baseline stress level was not associated with the amphetamine-induced increase in positive symptoms ($r = .05$, $p = .78$).

Effect of Gender

Ten women (four control subjects and six patients) were included in the study. Female patients displayed larger amphetamine-induced [^{123}I]IBZM displacement ($15.2 \pm 6.4\%$) compared to female control subjects ($5.2 \pm 5.3\%$, $p = .032$). No gender by diagnosis interaction was noted (dependent: [^{123}I]IBZM displacement; gender factor: $p = .49$; diagnosis factor: $p = .001$; gender by diagnosis interaction: $p = .97$). Therefore, the dysregulation in dopamine transmission revealed by the amphetamine-induced [^{123}I]IBZM displacement appeared to be present in patients with schizophrenia of both genders.

Effect of Previous Medication

Twenty seven patients were chronic and previously exposed to antipsychotic drugs, while 7 patients were in their first episode of illness and antipsychotic naive. The amphetamine effect on [^{123}I]IBZM binding potential was similar between the two groups (chronic/previously treated patients: $16.2 \pm 13.5\%$, $n = 27$; first episode/neuroleptic naive patients: $20.9 \pm 12.2\%$, $n = 7$, $p = .41$), and both groups were significantly different from control subjects (Figure 5, Table 3). In the previously treated group, the average time off neuroleptics prior to the scan was 104 ± 118 days (range 21 days to 360 days, with the latter value being used as index value for patients neuroleptic free for more than 1 year). No association was found between the duration of the neuroleptic-free period and the amphetamine-induced [^{123}I]IBZM displacement ($r = .02$, $p = .91$). Furthermore, the number of years of neuroleptic exposure was not associated with the amphetamine effect ($r = .14$, $p = .48$). Together, these data indicated that the exaggerated dopaminergic response

Table 3. Results: Comparison of Control Subjects; Chronic, Previously Treated Patients; and First-Episode, Neuroleptic-Naive Patients

Parameters	Control Subjects	Chronic, Previously Treated Patients	First-Episode, Neuroleptic-Naive Patients	*p*
n	36	27	7	—
Age	40 ± 9	42 ± 7	29 ± 9	<.01
Amphetamine-induced relative decrease in [^{123}I]IBZM BP (% baseline)	7.5% ± 7.1%	16.2% ± 13.5%	20.9% ± 12.2%	<.01

to amphetamine exposure was not a prolonged side effect of previous neuroleptic exposure.

Twelve patients received the benzodiazepine lorazepam p.o. (up to 3 mg q.d.) during the withdrawal phase, up to 24 hours prior to the scan. No effect of lorazepam on the dopaminergic response was detected, since amphetamine-induced [^{123}I]IBZM displacement was similar between the patients who did (17.1 ± 12.1%, $n = 12$) and did not (17.2% ± 14, $n = 22$, $p = .99$) receive lorazepam during the washout period.

Clinical Predictors of Dopaminergic Response

We tested associations between the amphetamine effect on [^{123}I]IBZM binding potential and several demographic and clinical variables in the patient group, in an attempt to characterize the profile of patients with exaggerated response. As already mentioned, the symptom severity (whether positive or negative symptoms) at baseline was not predictive of the amphetamine effect on D_2 receptor transmission. No association was found between the amphetamine effect and age ($r = .2$, $p = .90$), gender ($p = .69$), race ($p = .15$), subject socioeconomic status ($r = .05$, $p = .78$), familial socioeconomic status ($r = .07$, $p = .69$), duration of illness ($r < .01$, $p = .99$), or number of previous hospitalizations ($r = .01$, $p = .91$).

However, patients who were experiencing an illness exacerbation (as identified by the fact that their admission was motivated by clinical reasons) presented a higher amphetamine-induced [^{123}I]IBZM displacement (23.7 ± 13.2%, $n = 17$) than patients who were in remission and recruited as outpatients (10.5 ± 9.7%, $n = 17$, $p = .002$, Table 4, Figure 6). Furthermore, amphetamine-induced [^{123}I]IBZM displacement in remitted patients (10.5 ± 9.7%, $n = 17$) was not statistically different from control subjects (7.5 ± 7.1%, $n = 36$, $p = .27$). At baseline, patients in exacerbation had more positive symptoms than patients in remission, but this difference was not significant (patients in exacerbation: 18.9 ± 6.5; patients in remission: 16.1 ± 5.8, $p = .19$). Similarly, the amphetamine-induced increase in positive symptoms was higher in patients in exacerbation (3.8 ± 6.0) compared to patients in remission (2.1 ± 3.9), but this difference was not significant ($p = .33$). Patients in exacerbation tended to score higher on the post-amphetamine PANSS positive subscale (22.7 ± 1) compared to patients in remission (18.2 ± 8, $p = .085$). No differences were observed in the self-reported anxiety levels between these two groups, either at baseline (patients in exacerbation: 4.0 ± 2.0; patients in remission: 4.4 ± 1.8, $p = .58$), or following amphetamine (patients in exacerbation: 6.4 ± 2.4; patients in remission: 6.6 ± 2.5, $p = .84$). Thus, the scan data (dopamine release) provided a better discrimination between patients in remission and exacerbation, compared with the clinical ratings.

Baseline D_2 Receptor Binding Potential

Baseline [^{123}I]IBZM binding potential was not different between control subjects and patients (Table 2). Since the baseline binding potential is affected by baseline levels of endogenous dopamine (Laruelle et al 1997a), it was interesting to test the existence of an association between baseline binding potential and amphetamine effect. More

Table 4. Results: Comparison of Control Subjects, Patients in Remission, and Patients in Exacerbation

Parameters	Control Subjects	Patients in Remission	Patients in Exacerbation	*p*
n	36	17	17	—
Age	40 ± 9	42 ± 7	36 ± 10	.19
Amphetamine-induced relative decrease in [^{123}I]IBZM BP (% baseline)	7.5% ± 7.1%	10.5% ± 9.7%	23.7% ± 13.2%	<.001

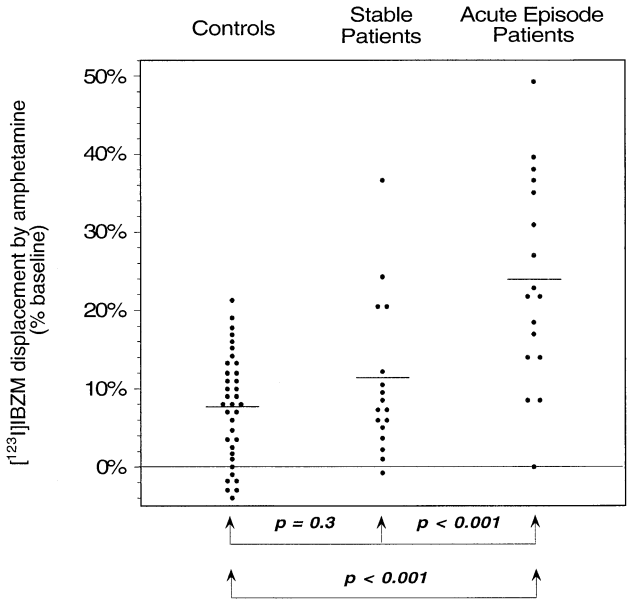

Figure 6. Amphetamine-induced reduction in [^{123}I]IBZM binding potential in healthy control subjects, patients with schizophrenia during a period of illness remission, and patients with schizophrenia during acute illness exacerbation.

specifically, we postulated that a high baseline binding potential could be associated with relatively lower levels of endogenous dopamine. In that case, the effective affinity for dopamine of the unoccupied D_2 receptors would be higher in subjects with high baseline binding potential value. Therefore, we tested the hypothesis that high baseline binding potential would be associated with high amphetamine effect. This correlation was indeed observed in control subjects, at an almost significant level ($r = .37$, $p = .056$). When controlled for age, this relationship between baseline binding potential and amphetamine effect became significant (two-way ANOVA, with baseline binding potential as dependent variable, age effect: $p = .01$, amphetamine effect: $p = .03$). Thus, in control subjects, we could postulate that a larger amphetamine effect was associated with low baseline endogenous dopamine levels, manifested by higher baseline D_2 receptor binding potential. This relationship may be mediated by an effective increased affinity of D_2 receptors due to lower competition by baseline dopamine levels.

Interestingly, this relationship was not detected in the schizophrenic group. Baseline [^{123}I]IBZM binding potential was not associated with amphetamine-induced [^{123}I]IBZM displacement ($r = .17$, $p = .40$). This lack of correlation was also present after correcting for age; in fact, age was not associated with a decrease in baseline [^{123}I]IBZM BP ($r = .26$, $p = .19$) in the schizophrenic subjects. This observation indirectly suggested that the increased effect of amphetamine in patients was not due to lower baseline dopamine levels compared to control subjects, and that the dysregulation of dopamine transmission revealed by the amphetamine challenge was associated with dysregulation of baseline dopamine activity. However, direct measurement of baseline dopamine levels will be needed to further explore the issue of the relationship between amphetamine-induced dopamine release and baseline (i.e., unchallenged) dopamine release (Laruelle et al 1997a).

Discussion

The analysis of this pooled and extended sample confirmed the results observed in the previous reports, and yielded interesting new results. The analysis confirmed that amphetamine-induced displacement of [^{123}I]IBZM specific binding is increased in schizophrenia, that a relatively large effect size of .95 is associated with this difference, that the within group variance of the amphetamine effect is increased in patients compared to control subjects, and that the excessive stimulation of dopamine transmission is significantly associated with worsening of positive symptoms. The new results derived from this analysis are as follows: 1) the stress associated with the procedure, although higher in patients than in control subjects does not appear to play a detectable role in the amphetamine-induced stimulation of dopamine transmission; 2) the dysregulation of dopamine transmission is present in both male and female patients; 3) the dysregulation is present in patients never previously exposed to neuroleptic drugs; 4) this dysregulation is present at onset of illness, and does not appear to worsen or improve with duration of illness; 5) this dysregulation is more pronounced during episodes of illness exacerbation, as opposed to periods of remission or stabilization; and 6) the baseline D_2 receptor availability explains some of the variance of the amphetamine effect in control subjects, but not in schizophrenic patients, indirectly suggesting an alteration in baseline dopamine levels in schizophrenia.

Effect Size

The effect size of the difference in the amphetamine-induced displacement between patients and control subjects (.95) is considerably larger than the effect size of other alterations described in schizophrenia using brain imaging techniques, such as alterations in ventricular or hippocampal size, or alteration of D_2 receptor density (Daniel et al 1991; Laruelle 1998; Lawrie and Abukmeil 1998; Nelson et al 1998). This finding supports the hypothesis that hyperresponsivity of dopaminergic neurons is an important component of the pathophysiology of schizophrenia. The interpretation of the scan data (increase

in [^{123}I]IBZM displacement) in terms of underlying mechanisms (increase in dopamine release) is supported by experimental data. In baboons, we previously established that the magnitude of amphetamine-induced reduction in [^{123}I]IBZM binding potential was correlated with the magnitude of the increase in extracellular dopamine measured with microdialysis (Laruelle et al 1997b), and similar results have been published by Breier and coworkers (1997). Thus, the increased displacement of [^{123}I]IBZM observed in the patient group is compatible with a larger increase in extracellular dopamine following amphetamine exposure in patients with schizophrenia compared to control subjects. Analysis of the baboon data yielded the following relationship between changes in extracellular dopamine (Δ dopamine, % of baseline) and reduction in [^{123}I]IBZM binding potential (Δ BP, % of baseline): Δ dopamine = 38.5Δ BP (assuming no intercept). Thus, an [^{123}I]IBZM binding potential reduction of 7.5% and 17.5% would correspond to an increase of 288% and 658% in extracellular dopamine in controls and patients, respectively.

Pre- Versus Post-Synaptic Mechanisms

Nevertheless, this interpretation rests on the assumption that the relationship between extracellular dopamine levels and D_2 receptor occupancy by dopamine is similar in patients and control subjects, i.e., that the affinity of D_2 receptors for dopamine is unchanged in schizophrenia. The sequence of the D_2 receptor gene, as well as the D_2 receptor affinity for antagonists is unaltered in schizophrenia (Gejman et al 1994; Seeman 1987). Moreover, the binding of dopamine agonists in postmortem striata is not increased in schizophrenia (Cross et al 1983; Lee et al 1978). Yet, the D_2 receptor affinity for agonists is regulated by multiple mechanisms such as coupling to G proteins (Sibley et al 1982), and we cannot rule out the possibility of a different in vivo affinity of dopamine for D_2 receptors in schizophrenia. Moreover, studies of D_1–D_2 receptor interactions in postmortem samples have suggested alterations of this interaction in schizophrenia, in a manner consistent with an increased affinity of D_2 receptors for dopamine in schizophrenia (Seeman et al 1989b). The development of radiolabeled D_2 receptor agonists as PET radiotracers is needed to address this issue. Until then, the interpretation of the results of this study in terms of increased dopamine levels remains tentative. In a strict sense, this study demonstrates that amphetamine leads to a greater stimulation of D_2 receptors in schizophrenia compared to control subjects, and the mechanism underlying this effect (increased dopamine concentration versus increased affinity of D_2 receptors for dopamine, or some combination of both factors) remains to be established.

The hypothesis that the observed increase in [^{123}I]IBZM displacement following amphetamine reflects mostly a dysregulation of dopaminergic neuron reactivity rather than a postsynaptic sensitivity, is indirectly supported by several recent investigations demonstrating an increase in [^{18}F]DOPA or [^{11}C]DOPA accumulation in the striatum of patients with schizophrenia (Hietala et al 1995; Reith et al 1994), but see Dao-Castellana and co-workers (1997). Together, these data suggest that an abnormality of D_2 receptor transmission is associated with schizophrenia, and that this abnormality results at least partly from an increase in presynaptic dopamine activity.

Limitations of the Competitive Model

It is also important to note that the interpretation of the amphetamine effect on [^{123}I]IBZM binding potential in terms of a simple competition model between dopamine and [^{123}I]IBZM does not account for all the data derived from preclinical experiments. For example, baboon studies have demonstrated that the duration of the effect of amphetamine on [^{123}I]IBZM binding potential exceeds the duration of the increase in extracellular dopamine measured with microdialysis, suggesting that a long-lasting adaptation of D_2 receptors to the surge in dopamine could play a role in the reduction in [^{123}I]IBZM binding potential. Furthermore, a simple competition model cannot explain the differences in the magnitude of amphetamine induced displacement of various D_2 radiotracers (Hartvig et al 1997). Differences in radiotracer affinity do not explain these differences in vulnerability to endogenous competition, either on a theorical or on an experimental basis (see discussion in Abi-Dargham et al 1999; Laruelle et al 1997b). Also, the observations that D_1 receptor agonists and antagonists are unaffected by dopamine competition are not easily explainable within the context of a simple competition model (Abi-Dargham et al 1999). Other mechanisms, such as receptor phosphorylation and internalization, and the impact of these regulations on radiotracer affinity, may play an important role in determining the effect of changes in endogenous dopamine levels on the in vivo binding of radiotracers (see discussion in Abi-Dargham et al 1999).

Irrespective of the exact mechanism driving the reduction in [^{123}I]IBZM binding potential following amphetamine, the blockade of this effect by dopamine depletion clearly establishes that dopamine release in a necessary first step in the cascade of events leading to the decrease in binding potential (Laruelle et al 1997b). Furthermore, the relationship between the magnitude of this effect and the amount of dopamine released in the extracellular space supports the usefulness of this measure to assess the intensity of D_2 receptor stimulation following this chal-

lenge. However, the remaining questions regarding the exact mechanism of this complex effect suggest that differences in endogenous dopamine levels may not be the only factors implicated in the differential response of [^{123}I]IBZM binding to amphetamine challenge between patients with schizophrenia and controls.

Striatal Dopamine and Positive Symptoms

This present pooled analysis clearly confirmed that the intensity of D_2 receptor stimulation by dopamine following amphetamine exposure explains a significant aspect of the variability in psychopathologic response to the challenge in schizophrenia. A number of studies, reviewed by Lieberman and colleagues (1987a) have provided evidence that schizophrenic patients, as a group, display increased sensitivity to the psychotogenic effects of acute psychostimulant administration. In other terms, some but not all patients with schizophrenia present emergence or worsening of psychotic symptoms after acute exposure to psychostimulants at doses that do not induce psychosis in healthy subjects. Key features of this response have emerged over the years, and were replicated in the present study. The clinical response to acute psychostimulant challenges in schizophrenic patients is highly heterogeneous. About 40% of patients present a worsening of positive psychotic symptoms, 40% show no change, and a minority (20%) do improve on positive symptoms following acute psychostimulant challenge (Lieberman et al 1987a; van Kammen et al 1982). The psychotogenic reaction is unrelated to the general behavioral activation (i.e., euphoria, restlessness, talkativeness) induced by the challenge. Thus, this reaction is more than a simple behavioral activation that would make the psychotic processes more observable (Angrist et al 1980; Janowsky and Davis 1976). When it occurs, the psychotic response is comparable to the "spontaneous" psychosis presented by the subject during the active episodes of the illness (Angrist et al 1980; Janowsky and Davis 1976; van Kammen et al 1982), a property that psychostimulants share with other psychotogenic agents, such as the NMDA antagonist ketamine (Lahti et al 1995).

In this sample, we observed a similar distribution of the quality of the response in patients with schizophrenia (47% worseners, 41% no change, and 12% improvers). Glucose metabolism changes following amphetamine challenge in schizophrenia do not characterize the features associated with a worsening of positive symptoms (Wolkin et al 1994). In contrast, this study provides evidence that amphetamine-induced dopamine release is an important determinant of this response. In other terms, the response is not uniquely characterized by differences in neuronal circuits "downstream" from the dopaminergic synapse. An exaggerated stimulation of D_2 receptors appears to be an important step for the expression of these symptoms. However, dopamine mediated stimulation of D_2 receptors explained only about 30% of the variance in the positive symptom change, indicating that other factors play a role in the exacerbation of these symptoms following amphetamine. Using the criteria of a 4-point increase in the positive subscale as a threshold for a significant emergence of positive symptoms, the regression of positive symptom change over [^{123}I]IBZM binding changes identifies a minimal decrease of 18.5% in [^{123}I]IBZM binding as the threshold associated with a clinically significant psychotic reaction. It is interesting to note that only 2 healthy control subjects exceeded this threshold, with displacement of 19% and 21%, respectively. These subjects did not present any detectable psychotic symptoms. On the other hand, 5 of the 16 patients classified as worseners displaced less than 18.5% of [^{123}I]IBZM specific binding, and 4 of the 18 patients classified as nonworseners displayed [^{123}I]IBZM displacement values higher than 18.5%. Thus, this threshold is not absolute, and other factors modulate the relationship between D_2 receptor activation and positive symptoms. The unique qualitative features of the psychotic response in each patient also support the idea that dopamine stimulation leads to activation of preexistent and specific dysfunctional neuronal circuits and reentrant ensembles that are specific to each patient. In other terms, acute dopamine stimulation might activate rather than create the neuronal activity associated with positive symptoms. This view is also consistent with the observation that sustained D_2 receptor blockade is necessary to allow the extinction of these putative dysfunctional ensembles. A differential sensitivity of these ensembles to dopamine stimulation between patients is consistent with the large variability in the response, which is not explainable by D_2 receptor activation.

We should also note that it is unclear if this increased responsivity of DA systems to amphetamine challenge is specific to schizophrenia, or represents a final common pathway for all conditions associated with psychotic symptoms responsive to D_2 receptor blockade (such as mania). Studies in nonschizophrenic patients with psychotic conditions are needed to clarify this issue.

Limitation in Anatomic Resolution

The dysfunctional neuronal reentrant loops supporting the experience of positive symptoms are likely to involve dysregulation of the prefrontal–ventral striatal–ventral pallidal–mediodorsal thalamic–prefrontal loop, and its regulation by hippocampal afferences (O'Donnell and Grace 1998). Considerable preclinical evidence from rodent

studies supports the hypothesis that antipsychotic drug action is associated with dopamine antagonism in the mesolimbic (ventral striatal, including nucleus accumbens) rather than the nigrostriatal (dorsostriatal) dopamine systems (for review see Deutch 1993). The limited resolution of the SPECT camera prevented us from distinguishing the respective contributions of the ventral and dorsal striata to the SPECT signal. The anterior striatum is a structure measuring an average of 20 to 25 mm along its longest axis in the coronal plane, with a ventral component (accumbens) of 6 to 8 mm and a dorsal component of 12 to 16 mm (Mai et al 1997). Thus, with SPECT (FWHM of 9 to 10 mm), the center of each structure is separated by only 1.5 FWHM, which does not allow separation. Based on average volumes of these structures, we estimated that only 15% to 20% of the striatal signal is derived from its ventral components. Thus, it is possible that the SPECT measurement considerably underestimates the magnitude of the dysregulation of DA release in schizophrenia if this dysregulation is more prominent in the ventral structures. On the other hand, the existence of a significant relationship between positive symptom emergence and increased dopamine transmission measured at the level of the whole striatum in this study, might suggest that the dopaminergic projections to the dorsal striatum also play a role in the pathophysiology of the positive symptoms (Lidsky 1995). Alternatively, this observation could indicate that an alteration in dopaminergic function measured at the level of the whole structure is predictive of the status of dopaminergic transmission in the ventral striatum. Studies performed with a high-resolution PET camera are needed to resolve this important issue.

Effect of Stress

Preclinical studies have shown that stress activates dopamine release (Deutch et al 1990; Finlay and Zigmond 1997; Kalivas and Duffy 1995), raising the question whether differences in procedure-induced stress between schizophrenics and control subjects may account for the differences observed in amphetamine-induced dopamine release. While subjects with schizophrenia reported higher levels of anxiety than control subjects prior and during the challenge, we failed to detect a correlation between dopamine release and anxiety level. Thus, stress did not appear to significantly contribute to the variability in amphetamine-induced dopamine release. The pharmacologic action of amphetamine on dopamine release may override the physiologic relationship between stress and dopamine release. Measurements of dopamine release under more physiologic conditions, such as during resting state (Laruelle et al 1997a) or during cognitive activation (Koepp et al 1998) would be more suitable for assessing the effect of stress on dopamine release. On the other hand, it is plausible that the dysregulation of dopamine transmission revealed by the amphetamine challenge in patients with schizophrenia could lead to excessive dopamine activity during stress, and mediate the relationship between stress and positive symptom exacerbation. The cross sensitization between amphetamine and environmental stressors on dopamine release supports the validity of the amphetamine challenge as a pharmacologic model of stress (Antelman 1980).

Striatal Dopamine and Negative Symptoms

Previous behavioral studies in patients with schizophrenia have shown that negative symptoms decrease or do not change following psychostimulant challenges (Angrist et al 1980), and that improvement in negative symptoms is predicted by their severity at baseline (Sanfilipo et al 1996). These observations were replicated in the present study. Amphetamine induced a significant reduction in the negative subscale of the PANSS, and the severity of baseline negative symptoms was predictive of the amphetamine-induced improvement on this scale. The observation of a significant relationship between increase in striatal dopamine transmission and improvement in negative symptoms is a new result that emerged from this combined and extended sample. This relationship, which did not reach significance in the two previously published cohorts (Abi-Dargham et al 1998; Laruelle et al 1996), became significant when data were combined, albeit to a lower degree than the relationship between excess dopamine transmission and positive symptoms. Furthermore, this correlation was essentially caused by two patients with both marked improvement in negative symptoms and marked amphetamine-induced dopamine release (Figure 3).

The association between an increase in dopamine transmission and improved negative symptoms is in agreement with the hypothesis that a deficiency in dopamine transmission is involved in the pathophysiology of negative symptoms. The concept of imbalance between a prefrontal cortical hypodopaminergic state underlying negative symptoms and a subcortical hyperdopaminergic state related to positive symptoms has been proposed to account for the coexistence, in schizophrenia, of both excess and deficiency of dopamine transmission (Davis et al 1991; O'Donnell and Grace 1998; Weinberger 1987). Our data do not contradict this hypothesis. The SPECT [^{123}I]IBZM method used here does not permit measurement of prefrontal dopamine release, given the very low number of D_2/D_3 receptors in the prefrontal cortex (Hall et al 1988). Studies with new PET radiotracers allowing assessment of DA transmission in extrastriatal areas will be needed to further explore this issue (Farde et al 1997; Mukherjee et

al 1997). Nevertheless, the observation that in two patients, a large reduction in negative symptoms was associated with a large amphetamine-induced decrease in [^{123}I]IBZM binding indirectly suggest that a hypodopaminergic state in the dorsal striatum could, in some patients, underlie negative symptoms.

Role of Previous Medications

In the previous two cohorts, we failed to detect a relationship between dopamine transmission response and duration of neuroleptic washout or lifetime neuroleptic exposure. This lack of correlation suggested that the observed effect was not a long-term side effect of previous neuroleptic exposure. However, this evidence was only circumstantial. In the present report, we present the results obtained in seven first-episode neuroleptic-free subjects. The amphetamine-induced dopamine release was similar, if not higher, in these subjects, indicating that the dysregulation of dopamine transmission revealed by the challenge is present at onset of illness and not due to previous medications. Similar data have been presented by Breier and co-workers (1997).

Relationship With Exacerbation of Illness

This extended analysis provided the opportunity to search for factors that may account for the variance of the effect in patients. In the two previously published cohorts, we failed to identify demographic or clinical features predictive of excessive dopamine release following amphetamine. Importantly, the severity of positive symptoms at baseline was not predictive of the dopaminergic response, and these results were confirmed in the present analysis. However, the present analysis revealed that the exaggerated dopamine response to amphetamine was mostly observed in the subset of patients who were studied during an episode of illness exacerbation, compared to patients in remission. These data suggest that the responsivity of the dopamine system might be predicted, not by the absolute level of baseline positive symptoms, but rather by the presence of an active phase of the illness. Since no clinical ratings were available for the time before the admission, we do not have direct evidence that patients classified as "in exacerbation" experienced a recent increase in symptomatology prior to admission. However, outpatient treatment and partial hospitalization were available treatment options for each of these patients and the decision was made that the clinical status required hospitalization. Under these conditions, hospitalization is one of the best indicators of "episode" and "relapse" (Schooler et al 1997), and it is reasonable to classify these patients as experiencing an active episode of the illness. A prospective study is warranted to further establish the relationship between dopamine dysregulation and illness exacerbation.

This observation is in agreement with previous studies reporting that the psychotic response to psychostimulants is state dependent in patients with schizophrenia. Patients who responded with a psychotic reaction to a psychostimulant challenge during an acute episode failed to show such a response when they were in remission (Janowsky and Davis 1976; van Kammen et al 1982). Furthermore, studies have shown that the vulnerability to psychostimulant-induced psychosis is associated with a higher rate of relapse upon neuroleptic discontinuation (Lieberman et al 1984; Lieberman et al 1987b). So, the propensity to present a psychotic reaction to a psychostimulant challenge may "reveal" an active phase of the illness not readily identifiable by the clinical symptomatology, in the absence of a psychostimulant challenge. The present study shows that a dysregulation of dopamine release could contribute to the pathophysiology of acute episodes. Remitted patients failed to show this abnormal response, also suggesting that the excess dopamine response is state dependent. Obviously, studying the same patients during exacerbation and remission is needed to confirm this point. However, the significance of the exacerbation/remission factor strongly suggests that increased dopamine release and psychosis exacerbation do not distinguish subgroups of patients, but rather different phases of the illness. This observation supports the idea that the symptomatic "heterogeneity" of schizophrenia could reside in the fluctuation of the illness over time, rather than in distinct subgroups of patients. Test/retest studies of amphetamine-induced dopamine release in healthy control subjects have shown that differences between subjects in the magnitude of this response are stable over time (Kegeles et al 1999), but this may not be true in patients with schizophrenia. This fluctuating nature of the dopaminergic abnormalities associated with schizophrenia should be considered when interpreting studies of genetic markers associated with this response. The absence of stable abnormality of dopamine function might also explain the inconsistency in detecting alterations in dopamine and its metabolites in postmortem studies of patients with schizophrenia (for review see Davis et al 1991).

Implication of Cortical-Subcortical Circuits

The mechanism of this increased dopaminergic neuronal reactivity remains unclear. The activity of dopaminergic cells in the ventral tegmental area (VTA) and substantia nigra (SN) is regulated, among others, by glutamatergic projections from the prefrontal cortex (Kalivas 1993; Karreman and Moghaddam 1996; Mathe et al 1998; Svensson and Tung 1989). This cortical glutamatergic

control occurs primarily through projections to the dopamine cell body area rather than the terminal region (Karreman and Moghaddam 1996). Given the evidence for dysfunction of the prefrontal cortex in schizophrenia, it is tempting to speculate that the dysregulation of subcortical dopamine revealed by this study might be secondary to a failure of the prefrontal, VTA/SN projections to properly regulate dopamine release following amphetamine challenge. Preclinical models have been proposed for such a dysregulation. In adult rats, a selective destruction of prefrontal dopamine terminals (Pycock et al 1980; Roberts et al 1994), an impairment of cortical GABAergic transmission (Karreman and Moghaddam 1996), or an impairment of NMDA transmission (Miller and Abercrombie 1996) can lead to a disinhibition of dopamine release. All three potential abnormalities have been proposed as models for this dysregulation in schizophrenia. Maybe more relevant, a neonatal lesion of the hippocampal formation leads to a failure of prefrontal–subcortical regulation of dopamine release in nonhuman primates (Saunders et al 1998), and this neurodevelopmental mechanism might offer a more realistic model of schizophrenia than acute manipulation in adults. However, the apparent fluctuation of dopamine function with the episode of illness is not fully accounted for by this neurodevelopmental model.

Exacerbation of Illness and Sensitization

Sensitization to psychostimulants is another well-described phenomenon in rodents that might be relevant to the disorders of dopamine function observed in this study (Lieberman et al 1997). Long-term sensitization to psychostimulants is a process whereby exposure to these drugs results in an enhanced response at subsequent exposures (for reviews see Kalivas and Stewart 1991; Kalivas et al 1993; Grace 1995; Robinson and Becker 1986). Several studies have shown that sensitization is associated with increased stimulant-induced dopamine release in the axonal terminal fields (Akimoto et al 1990; Kalivas and Duffy 1990; Kazahaya et al 1989; Patrick et al 1991; Paulson and Robinson 1995; Pettit et al 1990; Robinson et al 1988). Thus, sensitization in rodents is a paradoxical positive feedback loop, in which dopamine activation leads to more dopamine activation. Our data provide support for the hypothesis that dysfunction of dopamine systems in schizophrenia results from an endogenous sensitization process, since both conditions are associated with increased release (Lieberman et al 1990; Lieberman et al 1997). However, our data also suggest the extinction of this sensitized state during illness remission. The sensitization model may apply to the mechanism of illness exacerbation. Under stressful conditions, a positive feedback loop leads to larger activation of dopamine systems, which become autonomous, i.e., independent of stress and self-perpetuating, until D_2 receptor blockade interrupts this reaction, progressively allowing for its extinction. Upon neuroleptic discontinuation, the brain becomes again vulnerable to the stress-induced reemergence of this endogenous sensitization process. The neurodevelopmental abnormalities of brain connectivity and integration associated with schizophrenia might facilitate the emergence of these episodes of endogenous sensitization episodes.

Implication for Relapse Prevention

This model supports the need for new relapse prevention strategies. Currently, pharmacologic “maintenance” during remission phases is based on dopaminergic D_2 receptor blockade. These treatments succeed at reducing the relapse risk, but might do so at the price of inducing an unnecessary functional hypodopaminergic state, a condition associated with significant adverse effects and lower quality of life. A better understanding of the neurobiologic mechanisms that trigger the episodic states of dopaminergic hyperactivity associated with illness exacerbation might lead to new relapse prevention strategies sparing D_2 receptor function. In other terms, the apparent normality of dopamine transmission during illness remission might be a more important finding of these studies than the dysregulation during illness exacerbation. The development of animal neurodevelopmental models that lower dopamine sensitization threshold might ultimately lead to the development of alternative and less impairing relapse prevention pharmacologic strategies.

Conclusion

In conclusion, the extended analysis presented in this paper revealed some important new aspects of these studies not observed previously. Specifically, the observation that dysregulation of dopamine transmission is present at onset of illness and during periods of exacerbation, yet not detectable during periods of remission, provides a better understanding of the relationships between dopamine dysfunction and symptomatology, and might inform both preclinical models and drug development strategies.

We thank the subjects who participated in these studies; Paul Hoffer, MD, John Seibyl, MD, and Ronald Van Heertum, MD, for the support of the Departments of Nuclear Medicine at Yale and Columbia; John Krystal, MD, Dennis Charney, MD, J. John Mann, MD, and Jack Gorman, MD for the support of the Department of Psychiatry at Yale and Columbia; Janine Rodenhiser, PhD, Christine Fingado, Richard Weiss, and Lynn Pantages-Torok for excellent technical assistance. We also acknowledge the support of the National Alliance for Research on Schizophrenia and Depression (NARSAD), the EJLB Foundation, the

National Institute of Mental Health (RO1MH54192-0, K02 MH01603-01), and the Veterans Administration.

This work was presented at the conference, "Schizophrenia: From Molecule to Public Policy," held in Santa Fe, New Mexico in October 1998. The conference was sponsored by the Society of Biological Psychiatry through an unrestricted educational grant provided by Eli Lilly and Company.

References

Abi-Dargham A, Gil R, Krystal J, Baldwin R, Seibyl J, Bowers M, et al (1998): Increased striatal dopamine transmission in schizophrenia: Confirmation in a second cohort. *Am J Psychiatry* 155:761–767.

Abi-Dargham A, Simpson N, Kegeles L, Parsey L, Hwang D-R, Anjilvel S, et al (1999): PET studies of binding competition between endogenous dopamine and the D1 radiotracer [11C]NNC 756. *Synapse* 32:93–109.

Akimoto K, Hamamura T, Kazahaya Y, Akiyama K, Otsuki S (1990): Enhanced extracellular dopamine level may be the fundamental neuropharmacological basis of cross-behavioral sensitization between methamphetamine and cocaine—An in vivo dialysis study in freely moving rats. *Brain Res* 507:344–346.

Angrist B, Rotrosen J, Gershon S (1980) Response to apomorphine, amphetamine and neuroleptics in schizophrenic subjects. *Psychopharmacology* (Berlin) 67:31–38.

Angrist B, van Kammen DP (1984) CNS stimulants as a tool in the study of schizophrenia. *Trends Neurosci* 7:388–390.

Antelman S (1980): Interchangeability of stress and amphetamine in sensitization. *Science* 207:329–331.

Booij J, Korn P, Linszen DH, vanRoyen EA (1997): Assessment of endogenous dopamine release by methylphenidate challenge using iodine-123 iodobenzamide single-photon emission tomography. *Eur J Nucl Med* 24:674–677.

Breier A, Su TP, Saunders R, Carson RE, Kolachana BS, deBartolomeis A, Weinberger DR, et al (1997): Schizophrenia is associated with elevated amphetamine-induced synaptic dopamine concentrations: Evidence from a novel positron emission tomography method. *Proc Natl Acad Sci U S A* 94:2569–2574.

Burt DR, Creese I, Snyder SS (1977): Antischizophrenic drugs: Chronic treatment elevates dopamine receptors binding in brain. *Science* 196:326–328.

Carlsson A (1988): The current status of the dopamine hypothesis of schizophrenia. *Neuropsychopharmacology* 1:179–186.

Carlsson A, Lindqvist M (1963): Effect of chlorpromazine or haloperidol on formation of 3-methoxytyramine and normetanephrine in mouse brain. *Acta Pharmacol Toxicol* 20:140–144.

Carson RE, Breier A, deBartolomeis A, Saunders RC, Su TP, Schmall B, et al (1997): Quantification of amphetamine-induced changes in [C-11]raclopride binding with continuous infusion. *J Cereb Blood Flow Metab* 17:437–447.

Creese I, Burt DR, Snyder SH (1976): Dopamine receptor binding predicts clinical and pharmacological potencies of antischizophrenic drugs. *Science* 19:481–483.

Cross AJ, Crow TJ, Ferrier IN, Johnstone EC, McCreadie RM, Owen F, et al (1983): Dopamine receptor changes in schizophrenia in relation to the disease process and movement disorder. *J Neural Transm* (suppl 18):265–272.

Crow TJ (1980): Molecular pathology of schizophrenia: More than one disease process? *Br Med J* 280:66–68.

Daniel DG, Goldberg TE, Gibbons RD, Weinberger DR (1991): Lack of a bimodal distribution of ventricular size in schizophrenia: A Gaussian mixture analysis of 1056 cases and controls. *Biol Psychiatry* 30:887–903.

Dao-Castellana MH, Paillere-Martinot ML, Hantraye P, Attar-Levy D, Remy P, Crouzel C, et al (1997): Presynaptic dopaminergic function in the striatum of schizophrenic patients. *Schizophr Res* 23:167–174.

Davis KL, Kahn RS, Ko G, Davidson M (1991): Dopamine in schizophrenia: A review and reconceptualization. *Am J Psychiatry* 148:1474–1486.

Deutch A, Clark WA, Roth RH (1990): Prefrontal cortical dopamine depletion enhances the responsiveness of the mesolimbic dopamine neurons to stress. *Brain Res* 521:311–315.

Deutch AY (1993): Prefrontal cortical dopamine systems and the elaboration of functional corticostriatal circuits: Implications for schizophrenia and Parkinson's disease. *J Neural Transm* 91:197–221.

Dewey SL, Smith GS, Logan J, Brodie JD, Fowler JS, Wolf AP (1993): Striatal binding of the PET ligand ^{11}C-raclopride is altered by drugs that modify synaptic dopamine levels. *Synapse* 13:350–356.

Farde L, Nordström AL, Wiesel FA, Pauli S, Halldin C, Sedvall G (1992): Positron emission tomography analysis of central D_1 and D_2 dopamine receptor occupancy in patients treated with classical neuroleptics and clozapine. *Arch Gen Psychiatry* 49:538–544.

Farde L, Suhara T, Nyberg S, Karlsson P, Nakashima Y, Hietala J, et al (1997): A PET study of [C-11]FLB 457 binding to extrastriatal D-2-dopamine receptors in healthy subjects and antipsychotic drug-treated patients. *Psychopharmacology* 133:396–404.

Farde L, Wiesel F, Stone-Elander S, Halldin C, Nordsröm AL, Hall H, et al (1990): D2 dopamine receptors in neuroleptic-naive schizophrenic patients. A positron emission tomography study with [^{11}C]raclopride. *Arch Gen Psychiatry* 47:213–219.

Finlay JM, Zigmond MJ (1997): The effects of stress on central dopaminergic neurons: Possible clinical implications. *Neurochem Res* 22:1387–1394.

Gandelman MS, Baldwin RM, Zoghbi SS, Zea-Ponce Y, Innis RB (1994): Evaluation of ultrafiltration for the free fraction determination of single photon emission computerized tomography (SPECT) radiotracers: β-CIT, IBF and iomazenil. *J Pharm Sci* 83:1014–1019.

Gejman PV, Ram A, Gelernter J, Friedman E, Cao Q, Pickar D, et al (1994): No structural mutation in the dopamine D2 receptor gene in alcoholism or schizophrenia. Analysis using denaturing gradient gel electrophoresis. *JAMA* 271:204–208.

Grace AA (1995): The tonic/phasic model of dopamine system regulation: Its relevance for understanding how stimulant abuse can alter basal ganglia function. *Drug Alcohol Depend* 37:111–129.

Hall H, Farde L, Sedvall G (1988): Human dopamine receptors

subtypes-in vitro binding analysis using [3H]SCH 23390 and [3H]raclopride. *J Neural Transm* 73:7–21.

Hartvig P, Torstenson R, Tedroff J, Watanabe Y, Fasth KJ, Bjurling P, et al (1997): Amphetamine effects on dopamine release and synthesis rate studied in the Rhesus monkey brain by positron emission tomography. *J Neural Transm* 104:329–339.

Hietala J, Syvalahti E (1996): Dopamine in schizophrenia. *Ann Med* 28:557–561.

Hietala J, Syvälahti E, Vuorio K, Nagren K, Lehikoinen P, Ruotsalainen U, et al (1994): Striatal D2 receptor characteristics in neuroleptic-naive schizophrenic patients studied with positron emission tomography. *Arch Gen Psychiatry* 51:116–123.

Hietala J, Syvalahti E, Vuorio K, Rakkolainen V, Bergman J, Haaparanta M, et al (1995): Presynaptic dopamine function in striatum of neuroleptic-naive schizophrenic patients. *Lancet* 346:1130–1131.

Hollingshead AB (1975): *Four Factor Index of Social Status.* New Haven, CT: Working paper published by the author.

Innis RB, Malison RT, Al-Tikriti M, Hoffer PB, Sybirska EH, Seibyl JP, et al (1992): Amphetamine-stimulated dopamine release competes in vivo for [^{123}I]IBZM binding to the D_2 receptor in non-human primates. *Synapse* 10:177–184.

Inoue O, Kobayashi K, Tsukada H, Itoh T, Langstrom B (1991): Difference in in vivo receptor binding between [3H]N-methylspiperone and [3H]raclopride in reserpine-treated mouse brain. *J Neur Transm Gen Sect* 85:1–10.

Janowsky DS, Davis JM (1976): Methylphenidate, dextroamphetamine and levamphetamine. Effects on schizophrenic symptoms. *Arch Gen Psychiatry* 33:304–308.

Kalivas P, Duffy P (1990): The effect of acute and daily cocaine treatment on extracellular dopamine in the nucleus accumbens. *Synapse* 5:48–58.

Kalivas PW (1993): Neurotransmitter regulation of dopamine neurons in the ventral tegumental area. *Brain Res Rev* 18:75–113.

Kalivas PW, Duffy P (1995): Selective activation of dopamine transmission in the shell of the nucleus accumbens by stress. *Brain Res* 675:325–328.

Kalivas PW, Sorg BA, Hooks MS (1993): The pharmacology and neural circuitry of sensitization to psychostimulants. *Behav Pharmacol* 4:315–334.

Kalivas PW, Stewart J (1991): Dopamine transmission in the initiation and expression of drug- and stress-induced sensitization of motor activity. *Brain Res Rev* 16:223–244.

Karreman M, Moghaddam B (1996): The prefrontal cortex regulates the basal release of dopamine in the limbic striatum: An effect mediated by ventral tegmental area. *J Neurochem* 66:589–598.

Kay SR, Fiszbein A, Opler LA (1987): The Positive and Negative Syndrome scale (PANSS) for schizophrenia. *Schizophr Bull* 13:261–276.

Kazahaya Y, Akimoto K, Saburo O (1989): Subchronic methamphetamine treatment enhances methamphetamine- or cocaine induced dopamine efflux in vivo. *Biol Psychiatry* 25:903–912.

Kegeles L, Zea-Ponce Y, Abi-Dargham A, Rodenhiser J, Wang T, Wess R, et al (1999): Stability of [123I]IBZM SPECT measurement of amphetamine-induced dopamine release in healthy humans. *Synapse* 31:302–308.

Koepp MJ, Gunn RN, Lawrence AD, Cunningham VJ, Dagher A, Jones T, et al (1998): Evidence for striatal dopamine release during a video game. *Nature* 393:266–268.

Köhler C, Fuxe K, Ross SB (1981): Regional in vivo binding of [^{3}H]N-propylnorapomorphine in the mouse brain. Evidence for labelling of central dopamine receptors. *Eur J Pharmacol* 72:397–402.

Lahti AC, Koffel B, LaPorte D, Tamminga CA (1995): Subanesthetic doses of ketamine stimulate psychosis in schizophrenia. *Neuropsychopharmacology* 13:9–19.

Laruelle M (1998): Imaging dopamine transmission in schizophrenia. A review and meta-analysis. *Q J Nucl Med* 42:211–221.

Laruelle M, Abi-Dargham A, van Dyck CH, Gil R, De Souza CD, Erdos J, et al (1996): Single photon emission computerized tomography imaging of amphetamine-induced dopamine release in drug free schizophrenic subjects. *Proc Natl Acad Sci U S A* 93:9235–9240.

Laruelle M, Abi-Dargham A, van Dyck CH, Rosenblatt W, Zea-Ponce Y, Zoghbi SS, et al (1995): SPECT imaging of striatal dopamine release after amphetamine challenge. *J Nucl Med* 36:1182–1190.

Laruelle M, DSouza CD, Baldwin RM, Abi-Dargham A, Kanes SJ, Fingado CL, et al (1997a): Imaging D-2 receptor occupancy by endogenous dopamine in humans. *Neuropsychopharmacology* 17:162–174.

Laruelle M, Iyer RN, Al-Tikriti MS, Zea-Ponce Y, Malison R, Zoghbi SS, et al (1997b): Microdialysis and SPECT measurements of amphetamine-induced dopamine release in nonhuman primates. *Synapse* 25:1–14.

Lawrie SM, Abukmeil SS (1998): Brain abnormality in schizophrenia. A systematic and quantitative review of volumetric magnetic resonance imaging studies. *Br J Psychiatry* 172: 110–120.

Lee T, Seeman P, Tourtelotte WW, Farley IJ, Hornykiewicz O (1978): Binding of ^{3}H-neuroleptics and ^{3}H-apomorphine in schizophrenic brains. *Nature* 274:897–900.

Lidsky TI (1995): Reevaluation of the mesolimbic hypothesis of antipsychotic drug action. *Schizophr Bull* 21:67–74.

Lieberman JA, Kane JM, Alvir J (1987a): Provocative tests with psychostimulant drugs in schizophrenia. *Psychopharmacology* 91:415–433.

Lieberman JA, Kane JM, Gadaleta D, Brenner R, Lesser MS, Kinon B (1984): Methylphenidate challenge as a predictor of relapse in schizophrenia. *Am J Psychiatry* 141:633–638.

Lieberman JA, Kane JM, Sarantakos S, Gadaleta D, Woerner M, Alvir J, et al (1987b): Prediction of relapse in schizophrenia. *Arch Gen Psychiatry* 44:597–603.

Lieberman JA, Kinon BL, Loebel AD (1990): Dopaminergic mechanisms in idiopathic and drug-induced psychoses. *Schizophr Bull* 16:97–110.

Lieberman JA, Sheitman BB, Kinon BJ (1997): Neurochemical sensitization in the pathophysiology of schizophrenia: Deficits and dysfunction in neuronal regulation and plasticity. *Neuropsychopharmacology* 17:205–229.

Logan J, Dewey SL, Wolf AP, Fowler JS, Brodie JD, Angrist B, et al (1991): Effects of endogenous dopamine on measures of

[^{18}F]N-methylspiroperidol binding in the basal ganglia: Comparison of simulations and experimental results from PET studies in baboons. *Synapse* 9:195–207.

Mai JC, Assheuer J, Paxinos G (1997): *Atlas of the Human Brain.* San Diego, CA: Academic Press.

Mathe JM, Nomikos GG, Schilstrom B, Svensson TH (1998): Non-NMDA excitatory amino acid receptors in the ventral tegmental area mediate systemic dizocilpine (MK-801) induced hyperlocomotion and dopamine release in the nucleus accumbens. *J Neurosci Res* 51:583–592.

Miller DW, Abercrombie ED (1996): Effects of MK-801 on spontaneous and amphetamine-stimulated dopamine release in striatum measured with in vivo microdialysis in awake rats. *Brain Res Bull* 40:57–62.

Mukherjee J, Yang ZY, Lew R, Brown T, Kronmal S, Cooper MD, et al (1997): Evaluation of D-amphetamine effects on the binding of dopamine D-2 receptor radioligand, F-18-fallypride in nonhuman primates using positron emission tomography. *Synapse* 27:1–13.

Nelson MD, Saykin AJ, Flashman LA, Riordan HJ (1998): Hippocampal volume reduction in schizophrenia as assessed by magnetic resonance imaging: A meta-analytic study. *Arch Gen Psychiatry* 55:433–440.

O'Donnell P, Grace AA (1998): Dysfunctions in multiple interrelated systems as the neurobiological bases of schizophrenic symptom clusters. *Schizophr Bull* 24:267–283.

Owen F, Cross AJ, Crow TJ, Longden A, Poulter M, Riley GJ (1978): Increased dopamine-receptor sensitivity in schizophrenia. *Lancet* ii:223–226.

Patrick SL, Thompson TL, Walker JM, Patrik RL (1991): Concomitant sensitization of amphetamine-induced behavioral stimulation and in vivo dopamine release from rat caudate nucleus. *Brain Res* 538:343–346.

Paulson PE, Robinson TE (1995): Amphetamine-induced time-dependent sensitization of dopamine neurotransmission in the dorsal and ventral striatum: A microdialysis study in behaving rats. *Synapse* 19:56–65.

Pettit HO, Pan HT, Parsons LH, Justice JBJ (1990): Extracellular concentration of cocaine and dopamine are enhanced during chronic cocaine administration. *J Neurochem* 55:798–804.

Pilowsky LS, Costa DC, Ell PJ, Verhoeff NPLG, Murray RM, Kerwin RW (1994): D2 dopamine receptor binding in the basal ganglia of antipsychotic-free schizophrenic patients. An I-123-IBZM single photon emission computerized tomography study. *Br J Psychiatry* 164:16–26.

Pycock CJ, Kerwin RW, Carter CJ (1980): Effect of lesion of cortical dopamine terminals on subcortical dopamine receptors in rats. *Nature* 286:74–77.

Reith J, Benkelfat C, Sherwin A, Yasuhara Y, Kuwabara H, Andermann F, et al (1994): Elevated dopa decarboxylase activity in living brain of patients with psychosis. *Proc Natl Acad Sci U S A* 91:11651–11654.

Roberts AC, Desalvia MA, Wilkinson LS, Collins P, Muir JL, Everitt BJ, et al (1994): 6-hydroxydopamine lesions of the prefrontal cortex in monkeys enhance performance on an analog of the Wisconsin card sort test: Possible interactions with subcortical dopamine. *J Neurosci* 14:2531–2544.

Robinson PH, Jurson PA, Bennet JA, Bentgen KM (1988): Persistent sensitization of dopamine neurotransmission in ventral striatum (nucleus accumbens) produced by prior experience with (+)-amphetamine: A microdialysis study in freely moving rats. *Brain Res* 462:211–222.

Robinson TE, Becker JB (1986): Enduring changes in brain and behavior produced by chronic amphetamine administration: A review and evaluation of animal models of amphetamine psychosis. *Brain Res Review* 11:157–198.

Ross SB (1991): Synaptic concentration of dopamine in the mouse striatum in relationship to the kinetic properties of the dopamine receptors and uptake mechanism. *J Neurochem* 56:22–29.

Ross SB, Jackson DM (1989a): Kinetic properties of the in vivo accumulation of the dopamine D-2 receptor antagonist raclopride in the mouse brain in vivo. *Naunyn Schmiedeberg Arch Pharmacol* 340:6–12.

Ross SB, Jackson DM (1989b): Kinetic properties of the in vivo accumulation of [^{3}H](−)-N-n-propylnorapomorphine in the mouse brain. *Naunyn Schmiedeberg Arch Pharmacol* 340:13–20.

Sanfilipo M, Wolkin A, Angrist B, van Kammen DP, Duncan E, Wieland S, et al (1996): Amphetamine and negative symptoms of schizophrenia. *Psychopharmacology* (Berlin) 123: 211–214.

Saunders RC, Kolachana BS, Bachevalier J, Weinberger DR (1998): Neonatal lesions of the medial temporal lobe disrupt prefrontal cortical regulation of striatal dopamine. *Nature* 393:169–171.

Schooler NR, Keith SJ, Severe JB, Matthews SM, Bellack AS, Glick ID, et al (1997): Relapse and rehospitalization during maintenance treatment of schizophrenia. The effects of dose reduction and family treatment. *Arch Gen Psychiatry* 54:453–463.

Seeman P (1987): Dopamine receptors and the dopamine hypothesis of schizophrenia. *Synapse* 1:133–152.

Seeman P, Bzowej NH, Guan HC, Bergeron C, Reynolds GP, Bird ED, Riederer P, et al (1987): Human brain D1 and D2 dopamine receptors in schizophrenia, Alzheimer's, Parkinson's, and Huntington's diseases. *Neuropsychopharmacology* 1:5–15.

Seeman P, Guan H-C, Niznik HB (1989a): Endogenous dopamine lowers the dopamine D_2 receptor density as measured by [^{3}H]raclopride: Implications for positron emission tomography of the human brain. *Synapse* 3:96–97.

Seeman P, Lee T (1975): Antipsychotic drugs: Direct correlation between clinical potency and presynaptic action on dopamine neurons. *Science* 188:1217–1219.

Seeman P, Niznik HB, Guan HC, Booth G, Ulpian C (1989b): Link between D1 and D2 dopamine receptors is reduced in schizophrenia and Huntington diseased brain. *Proc Natl Acad Sci U S A* 86:10156–10160.

Sibley DR, De Lean A, Creese I (1982): Anterior pituitary receptors: Demonstration of interconvertible high and low affinity states of the D_2 dopamine receptor. *J Biol Chem* 257:6351–6361.

Svensson TH, Tung CS (1989): Local cooling of pre-frontal cortex induces pace-maker-like firing of dopamine neurons in rat ventral tegumental area in vivo. *Acta Physiol Scand* 136:135–136.

Van der Werf JF, Sebens JB, Vaalburg W, Korf J (1986): In vivo

binding of N-n-propylnorapomorphine in the rat brain: Regional localization, quantification in striatum and lack of correlation with dopamine metabolism. *Eur J Pharmacol* 87:259–270.

van Kammen DP, Bunney WE, Jr., Docherty JP, Marder SR, Ebert MH, Rosenblatt JE, et al (1982): D-Amphetamine-induced heterogeneous changes in psychotic behavior in schizophrenia. *Am J Psychiatry* 139:991–997.

van Kammen DP, Murphy DL (1975): Attenuation of the euphoriant and activating effects of D- and L-amphetamine by lithium carbonate treatment. *Psychopharmacologia* (Berlin) 44:215–224.

Volkow ND, Wang G-J, Fowler JS, Logan J, Schlyer D, Hitzemann R, et al (1994): Imaging endogenous dopamine competition with [^{11}C]raclopride in the human brain. *Synapse* 16:255–262.

Weinberger DR (1987): Implications of the normal brain development for the pathogenesis of schizophrenia. *Arch Gen Psychiatry* 44:660–669.

Willner P (1997): The dopamine hypothesis of schizophrenia: Current status, future prospects. *Int Clin Psychopharmacol* 12:297–308.

Wolkin A, Sanfilipo M, Angrist B, Duncan E, Wieland S, Wolf AP, et al (1994): Acute D-amphetamine challenge in schizophrenia: Effects on cerebral glucose utilization and clinical symptomatology. *Biol Psychiatry* 36:317–325.

Wong DF, Wagner HN, Tune LE, Dannals RF, Pearlson GD, Links JM, et al (1986): Positron emission tomography reveals elevated D_2 dopamine receptors in drug-naive schizophrenics. *Science* 234:1558–1563.

Young LT, Wong DF, Goldman S, Minkin E, Chen C, Matsumura K, et al (1991): Effects of endogenous dopamine on kinetics of [^{3}H]methylspiperone and [^{3}H]raclopride binding in the rat brain. *Synapse* 9:188–194.